Feasibility Study for a Compact, Multi-Purpose Bioluminescence Detector

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LONG-TERM GOALS

My research objectives involve determining how low light phenomena, both bioluminescence and solar radiation below 200 meters, influence the distribution and behavior of marine organisms.

OBJECTIVES

The objective is to test the feasibility of developing a low-cost, compact bioluminescence detector based on the design principles used to develop the High Intake Defined Excitation Bathyphotometers (HIDEX-BP) (Widder et al., 1993). Specifically the design requirements are: 1) defined excitation in order to quantify the stimulus, 2) high flow rates in order to improve sampling statistics, and 3) a long residence time capable of measuring an entire flash.

APPROACH

The basic design of the HIDEX-BP is such that bioluminescence is stimulated by hydrodynamically calibrated flow through a turbulence generating grid at the entrance to a large cylindrical detection chamber. An array of optical fibers, embedded in the walls of the detection chamber, collect light and direct it to a photomultiplier tube. Two of the high costs associated with this design are: 1) the high speed submersible pump and 2) the machining costs associated with embedding the fiber optics in the walls of the detection chamber. The approach has been to reduce these costs and simplify the design by using off-the-shelf components and to scale down the size to the minimum needed to maintain HIDEX-BP design principles.

WORK COMPLETED

The mini-HIDEX has been constructed and is currently undergoing tests using laboratory cultures of bioluminescent dinoflagellates to calibrate the system biologically. The spatial distribution of stimulated bioluminescence in the detection chamber has been recorded using an intensified video camera viewing a full scale Plexiglas model of the mini-HIDEX. The throughput and field-of-view of the 3.5 in. light ring detector system has been measured and the field-of-view was found to be a good match for the spatial distribution of stimulated bioluminescence. Based on the throughput measurements a PMT light detector module was selected over the alternative silicon photodiode with built-in op amp that was being considered for this application. The light baffle/flow meter was constructed and a low cost, high-speed submersible pump, which met design specifications, was located and purchased. An integrating sphere system was designed and calibrated for the biological

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calibrations. As soon as the biological calibrations are complete the system will be set up for long-term data collection in a moored configuration off the HBOI sea wall.

RESULTS

Results so far indicate that this low-cost compact system may be a viable alternative to the HIDEX-BP. The primary limitation seems to be the submersible pumps. The pump which we have located appears to be a good solution for shallow water environments but does not have an adequate depth rating for a profiling system. The critical importance of a high-speed pump that can capture fast swimming zooplankton, which may evade slower pumping systems, has been made apparent by our recent discovery of thin layers of bioluminescent copepods in the Gulf of Maine (Widder and Johnsen, 1998a; Widder and Johnsen, in press; Widder et al., submitted). These layers were discovered using the HIDEX-BP operated at a pump speed of 16 L/sec. The slower pumping rate of the mini-HIDEX (4 L/sec) should be adequate for most coastal bioluminescence, which is likely to be primarily dinoflagellates, ostracods and gelatinous zooplankton, however a faster pump speed would be desirable in a profiling system. With this in mind we have made the design modular so that it can be adapted to other pump and/or thruster designs. Additionally we have designed the light-ring detector system so that it can be adapted to our bioluminescence imaging system (see Related Projects below).

IMPACT

There is very little data available on bioluminescence potential in coastal zones. Because of the extreme variability in these environments, extensive sampling is needed in order develop some degree of predictive capability. The development of low-cost, compact bioluminescence detectors, which can be assembled from largely off-the-shelf components, could significantly increase the database on coastal zone bioluminescence. This would benefit both civilian and defense sectors by providing a valuable tool for biological oceanographers interested in plankton distribution patterns as well as providing standardized measurements for calculating the impact which bioluminescence may have on covert naval operations. Our results indicate that this design is feasible and a highly desirable alternative to the detector systems still in use, which are limited by low-flow, an undefined stimulus regime and short residence times.

TRANSITIONS

Conclusions from this study will be made available through a publication in the open literature.

RELATED PROJECTS

The design of the light ring detector system can be adapted to our on going efforts to develop an *in situ* video transect technique that uses the unique temporal and spatial characteristics of bioluminescent displays to identify sources. Using an intensified video camera to image bioluminescence in situ we have been developing a database of identified bioluminescent signatures (Widder et al., 1989; Widder et al., 1992; Widder et al., 1997). We have also been developing image recognition programs that can identify sources based on their signatures (Kocak et al., in press; Widder and Johnsen, 1998; Widder and Johnsen, in press; Widder, et al., in press). The light

ring detector module can be used in combination with the imaging system to provide the high-resolution intensity and kinetics data, which are the critical to accurate identifications. In another related project, accurate calibration of this unusual measurement geometry has been made possible with the development of the light wand calibration source developed in our laboratory with ONR support (see HIDEX-BP Calibrations report).

REFERENCES

Kocak, D.M., N.D.V. Lobo, and E.A. Widder. (in press) Computer vision techniques for quantifying, tracking and identifying bioluminescent plankton. IEEE J. Oceanic Engineering.

Widder, E.A. (1997) *In situ* video recordings of bioluminescence in the ostracod, *Conchoecia elegans*, and co-occuring bioluminescent zooplankton in the Gulf of Maine. In: Proceedings of the 9th International Symposium on Bioluminescence and Chemiluminescence. Eds. JW Hastings, LJ Kricka and PE Stanley. John Wiley & Sons Ltd, Sussex, UK. pp. 159-164.

Widder, E.A., S. Bernstein, D. Bracher, J.F. Case, K.R. Reisenbichler, J.J. Torres and B.H. Robison. (1989) Bioluminescence in Monterey Submarine Canyon: image analysis of video recordings from a midwater submersible. Mar. Biol. 100:541-551.

Widder, E.A., J.F. Case, S.A. Bernstein, S. MacIntyre, M.R. Lowenstine, M.R. Bowlby, and D.P. Cook. (1993) A new large volume bioluminescence bathyphotometer with defined turbulence excitation. Deep Sea Res. 40(3): 607-627.

Widder, E.A., C.H. Greene and M.J. Youngbluth. (1992) Bioluminescence of sound-scattering layers in the Gulf of Maine. J. Plank. Res. 14(11): 1607-1624.

Widder, E.A., and S. Johnsen (1998) Spatial Plankton Analysis Technique (SPLAT) for 3D Reconstruction and Statistical Analysis of Bioluminescent Plankton Spatial Point Patterns. Eos Trans. Amer. Geophys. Union. 79(1):OS51G-6

Widder, E.A. and S. Johnsen. (in press) Optical imaging, identification and 3D analysis of spatial distribution patterns of bioluminescent plankton. SPIE Ocean Optics XIV.

Widder, E.A., S. Johnsen, S. A. Bernstein, J. F. Case, D. J. Neilson. (submitted) Thin layers of bioluminescent copepods found at density discontinuities in the water column.

PUBLICATIONS

Widder, E.A. (1997) *In situ* video recordings of bioluminescence in the ostracod, *Conchoecia elegans*, and co-occuring bioluminescent zooplankton in the Gulf of Maine. In: Proceedings of the 9th International Symposium on Bioluminescence and Chemiluminescence. Eds. JW Hastings, LJ Kricka and PE Stanley. John Wiley & Sons Ltd, Sussex, UK. pp. 159-164.

Makemson, J.C., N.R. Fulayfil, W.L. Landry, L.M. Van Ert, C.F. Wimpee, E.A. Widder and J.F. Case (1997) *Shewanella woodyi* sp. nov., an exclusively respiratory luminous bacterium isolated from the Alboran Sea. International Journal of Systematic Bacteriology 47 (4): 1034-1039.

Frank, T.M. and E.A. Widder (1997) The correlation of downwelling irradiance and staggered vertical migration patterns of zooplankton in Wilkinson Basin, Gulf of Maine. J. Plankton Research. 19 (12): 1975-1991.

Haddock, S.H.D. Neilson, D.J. Widder, E.A. and Case, J.F. (1998) Feasibility of using in situ measurements of bioluminescence spectra to determine the vertical distribution of plankton. In: Pelagic Biogeography IcoPB II Proceedings of the 2nd International Conference Workshop Report 142:137-142.

Widder, E.A. (1998) A predatory use of counterillumination by the squaloid shark, *Isistius brasiliensis*. Environmental Biology of Fishes. 53:267-273.

Widder, E. A., and T.M. Frank (1998) The Speed of an Isolume: A Shrimp's Eye View. Eos Trans. Amer. Geophys. Union. 79(1):OS21E-2

Frank, T.M. and E.A. Widder (1998) In situ measurements of the distribution patterns of vertical migrators in Oceanographer Canyon correlated with in situ measurements of downwelling light. Eos Trans. Amer. Geophys. Union. 79(1):OS42H-11

Widder, E.A., S. Johnsen (1998) Spatial Plankton Analysis Technique (SPLAT) for 3D Reconstruction and Statistical Analysis of Bioluminescent Plankton Spatial Point Patterns. Eos Trans. Amer. Geophys. Union. 79(1):OS51G-6

Widder, E.A. (1998) Bioluminescence. In: "Adaptive Mechanisms in the Ecology of Vision." Edited by: S.N. Archer, M.B.A. Djamgoz, E. Loew, J.C. Partridge & S. Vallerga. Chapman & Hall, London. (in press)

Johnsen, S. and E.A. Widder. (1998) Measurements of the transparency of mesopelagic gelatinous zooplankton from the North Western Atlantic. Biol. Bull. (in press).

Kocak, D.M., N.D.V. Lobo, and E.A. Widder. (1998) Computer vision techniques for quantifying, tracking and identifying Bioluminescent plankton. IEEE J. Oceanic Engineering. (in press)

Widder, E.A. and S. Johnsen. (1998) Optical imaging, identification and 3D analysis of spatial distribution patterns of bioluminescent plankton. SPIE Ocean Optics XIV. (in press)

Johnsen, S. and E.A. Widder. (1998) The transparency and visibility of gelatinous zooplankton. SPIE Ocean Optics XIV. (in press)

Johnsen, S. and E.A. Widder. (submitted) The physical basis of transparency in biological tissue: Ultrastructure and the minimization of light scattering.

Bohnsack, J.A. and E.A. Widder. (submitted) The potential of optical technology for advancing marine resource assessment.

Widder, E.A., S. Johnsen, S. A. Bernstein, J. F. Case, D. J. Neilson. (submitted) Thin layers of bioluminescent copepods found at density discontinuities in the water column.

Johnsen, S., E.J. Balser and E.A. Widder. (submitted) Modified suckers as light organs in a deep-sea octopod.